Amendments To The Claims

This listing of claims will replace all prior versions, and listings, of claims in the application:

Listing of Claims

Claim 1 (Currently Amended): A method for ~~determining the rate of transcription of~~ quantifying nascent RNA transcripts from a transcriptional unit in a preparation of nuclei ~~a composition~~ of cells, said method comprising:

~~lysing the cells and obtaining from the cells~~ providing a preparation of isolated cell nuclei comprising said transcriptional unit with nascent RNA transcripts ~~strands~~ attached thereto, wherein transcription of the transcriptional unit in said nuclei has been reversibly inhibited ~~and placing same on ice to temporarily inhibit continued transcription and then~~

placing said nuclei under conditions to permit transcription of the transcriptional unit in the presence of a ribonucleotide with a purifyable label ~~biotin-16-UTP, wherein said biotin-16-UTP includes a cleavable linker between the biotin and the UTP~~, to thereby provide a population of ~~biotin~~ purifyably-labeled nascent RNA transcripts; and

isolating said ~~biotin~~ purifyably-labeled nascent RNA transcripts by immobilizing said nascent RNA transcripts on a matrix having affinity for said label and subsequently releasing said nascent RNA transcripts from said matrix ~~same onto streptavidin-labeled iron beads purifying said RNA transcripts by magnetic separation, cleaving said biotin-16-UTP at said cleavable linker after said magnetic separation to thereby provide a population of nascent RNA transcripts~~, and

quantitatively determining the level of the nascent RNA transcripts ~~by subjecting the RNA transcripts to real-time PCR~~.

Claim 2 (Currently Amended): The method of Claim 1 wherein the isolated cells nuclei are mammalian nuclei ~~cells~~.

Claims 3-5 (Canceled).

Claim 6 (Currently Amended): A method for ~~determining the rate of transcription of~~ quantifying nascent RNA transcripts from one or more transcriptional units in one or more cells, said method comprising:

obtaining from said one or more cells a preparation of nucleic acids comprising said one or more transcriptional units with nascent RNA strands attached thereto,

inhibiting continued transcription of said nucleic acids,

placing said nucleic acids under conditions to permit transcription of said transcriptional unit in the presence of ~~biotin~~ purifiably-labeled ribonucleotides, ~~wherein said biotin-labeled ribonucleotides include a cleavable linker between said biotin and said ribonucleotide,~~ to thereby provide a population of ~~biotin~~ purifiably-labeled nascent RNA transcripts ~~that include a cleavable linker between said biotin and said ribonucleotide~~;

isolating said ~~biotin~~ purifiably-labeled nascent RNA transcripts by immobilizing said label onto a solid matrix having affinity for said label,

~~cleaving said biotin-labeled ribonucleotide at said cleavable linker~~ releasing said nascent RNA transcripts from said matrix to thereby provide a population of nascent RNA transcripts;

and ~~subjecting said nascent RNA transcripts to a real-time polymerase chain reaction to determine the rate of transcription of~~ quantifying the nascent RNA transcripts from said one or more transcriptional units.

Claim 7 (Currently Amended): The method of claim 6, wherein the purifyably-labeled ribonucleotides are biotin-labeled ribonucleotides with a cleavable linker between said biotin and said ribonucleotides ~~cleavable linker is a disulfide S-S bridge~~.

Claims 8-9 (Canceled).

Claim 10 (Previously Presented): The method of claim 6 wherein said preparation of nucleic acids comprises cellular or viral nucleic acids.

Claim 11 (Previously Presented): The method of claim 6 wherein said nucleic acids are from nuclei, mitochondria, or chloroplasts.

Claim 12 (Previously Presented): The method of claim 11 wherein said nucleic acids are from nuclei.

Claim 13 (Previously Presented): The method of claim 6 comprising the additional step of purifying said biotin-labeled nascent RNA transcripts.

Claim 14 (Previously Presented): The method of claim 6 wherein said inhibiting is accomplished by cooling said nucleic acids.

Claim 15 (Previously Presented): The method of claim 6 wherein said one or more cells are mammalian cells.

Claim 16 (Currently Amended): The method of claim ~~6~~ 7 wherein said biotin-labeled ribonucleotides are biotin-16-UTP.

Claim 17 (Canceled).

Claim 18 (Currently Amended): A method of detecting a change in quantifiable nascent RNA transcripts from ~~activity of~~ one or more transcriptional units, comprising:

~~determining the rate of transcription in~~ quantifying amounts of nascent RNA transcripts from one or more transcriptional units of a first portion of cells according to the method of claim 6,

exposing a second portion of cells to one or more internal or external stimuli,

~~determining the rate of transcription in~~ quantifying amounts of nascent RNA transcripts from one or transcriptional units of said second portion of said cells according to the method of claim 6, and

comparing said amounts of nascent RNA transcripts from ~~rate of transcription of~~ said one or more transcriptional units of said first portion and amounts of nascent RNA transcripts from said one or more transcriptional units of said second portion to detect a change in quantifiable nascent RNA transcripts from ~~activity of~~ said one or more transcriptional units.

Claim 19 (Previously Presented): A method of claim 18, wherein said one or more internal or external stimuli comprise at least a portion of the nucleic acid sequence of an endogenous gene.

Claim 20 (Previously Presented): A method of claim 18, wherein said one or more internal or external stimuli is an exogenous transgene.

Claim 21 (Currently Amended): A method of detecting a change in quantifiable nascent RNA transcripts from ~~activity of~~ one or more transcriptional units at different stages of cellular development, comprising:

~~determining the rate of transcription in~~ quantifying amounts of nascent RNA transcripts from said one or more transcriptional units according to the method of claim 6 at two or more different stages of cellular development, and

comparing said ~~rates of transcription~~ amounts of nascent RNA transcripts at said two or more stages of development to detect a change in quantifiable nascent RNA transcripts from ~~activity of~~ said one or more transcriptional units.

Claims 22-29 (Canceled).

Claim 30 (Currently Amended): A method for ~~determining the rate of transcription of~~ quantifying nascent RNA transcripts from one or more transcriptional units in one or more cells, said method comprising:

obtaining from said one or more cells a preparation of nucleic acids comprising said one or more transcriptional units with nascent RNA strands attached thereto,

inhibiting continued transcription of said nucleic acids,

placing said nucleic acids under conditions to permit transcription of said transcriptional unit in the presence of biotin-labeled ribonucleotides to thereby provide a population of biotin-labeled nascent RNA transcripts;

isolating said biotin-labeled nascent transcripts by immobilizing said label onto a solid matrix, wherein said matrix includes a cleavable linker;

cleaving said matrix at said cleavable linker to thereby provide a population of biotin-labeled nascent RNA transcripts;

and ~~subjecting said biotin-labeled nascent RNA transcripts to a real-time polymerase chain reaction to determine the rate of transcription of~~ quantifying the nascent RNA transcripts from said one or more transcriptional units.

Claim 31 (Previously Presented): The method of claim 30, wherein the cleavable linker is a disulfide S-S bridge.

Claim 32 (Canceled).